

Cab391 Cas9-CKO Strategy

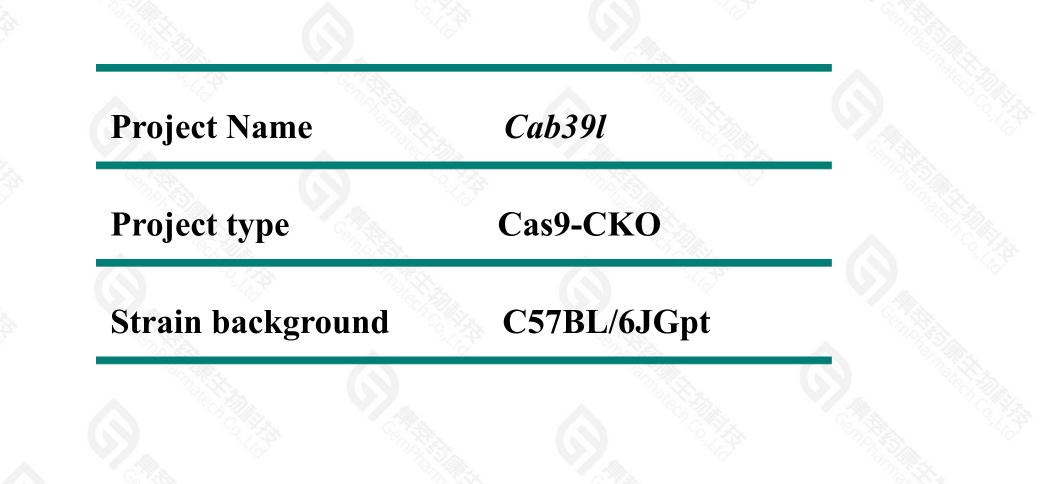
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Reviewer: Miaomiao Cui

Design Date: 2021-4-23

Project Overview





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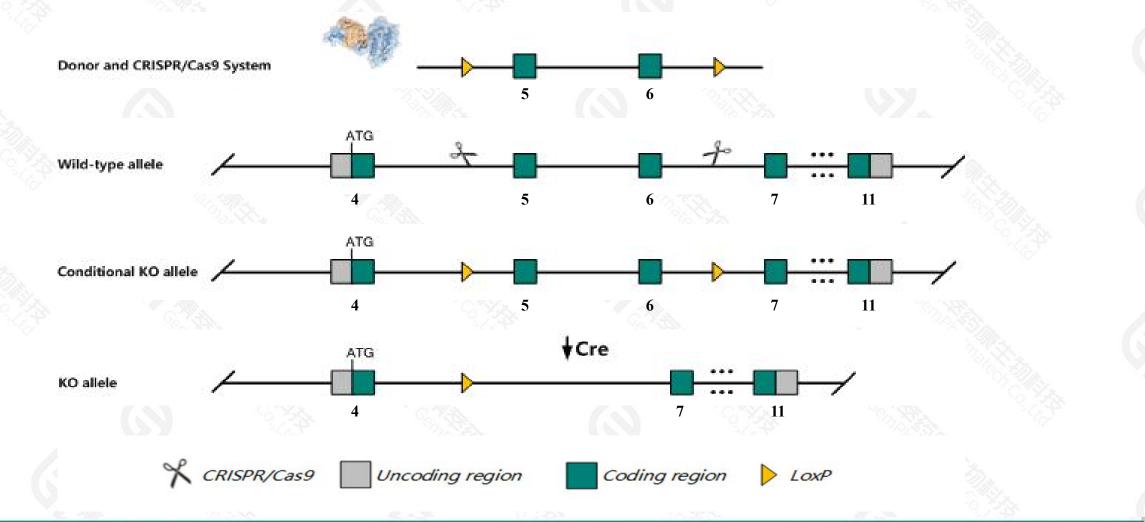
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Conditional Knockout strategy

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This model will use CRISPR/Cas9 technology to edit the Cab39l gene. The schematic diagram is as follows:



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Technical routes



The Cab391 gene has 6 transcripts. According to the structure of Cab391 gene, exon5-exon6 of Cab391-201(ENSMUST0000022553.6) transcript is recommended as the knockout region. The region contains 284bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Cab39l* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

> The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.



➤ Transcript *Cab391*-204 may not be affected.

> The *Cab391* gene is located on the Chr14. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.

> This strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes,all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

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Gene information (NCBI)

Cab39I calcium binding protein 39-like [Mus musculus (house mouse)]

Gene ID: 69008, updated on 17-Dec-2020

Summary

Official Symbol	Cab39I provided by MGI
Official Full Name	calcium binding protein 39-like provided by MGI
Primary source	MGI:MGI:1914081
See related	Ensembl:ENSMUSG0000021981
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;
	Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1500031K13Rik, 2810425013Rik, 4930520C08Rik, AA589432, MO, MO2L
Expression	Ubiquitous expression in bladder adult (RPKM 15.1), testis adult (RPKM 7.4) and 28 other tissuesSee more
Orthologs	human all



☆ ?

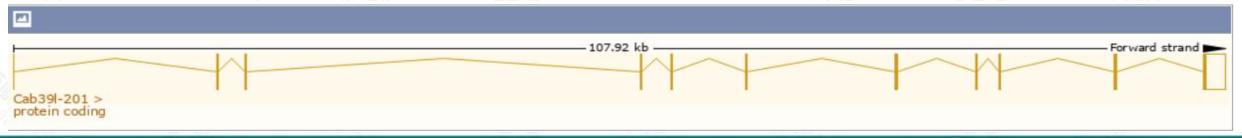
Transcript information (Ensembl)

The gene has 6 transcripts, all transcripts are shown below:

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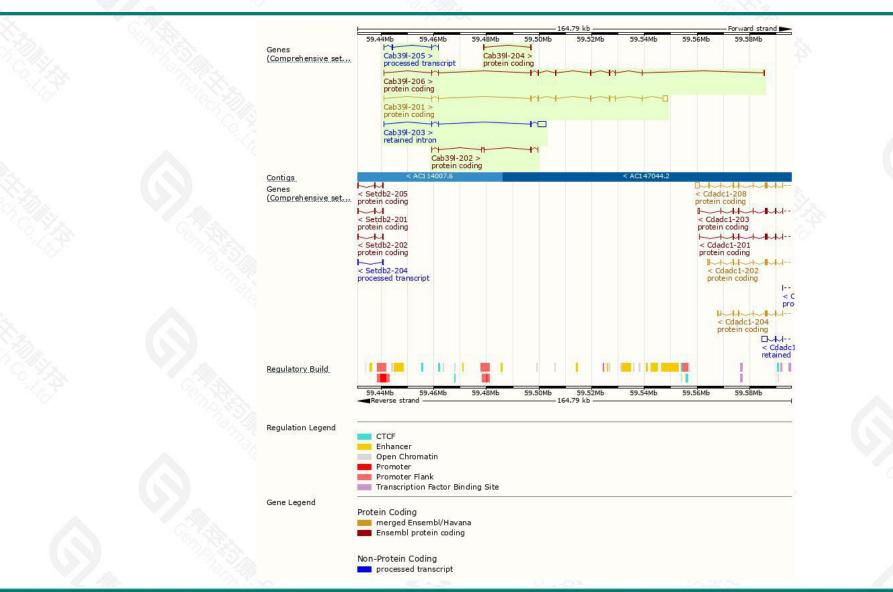
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cab39l-201	ENSMUST0000022553.6	3037	<u>337aa</u>	Protein coding	CCD527171		TSL:1, GENCODE basic, APPRIS P1,
Cab39l-206	ENSMUST00000225595.2	1380	<u>296aa</u>	Protein coding	-		GENCODE basic ,
Cab39l-202	ENSMUST00000223678.2	623	<u>79aa</u>	Protein coding	2		CDS 3' incomplete ,
Cab39l-204	ENSMUST00000224893.2	496	<u>32aa</u>	Protein coding			CDS 3' incomplete ,
Cab39l-205	ENSMUST00000225149.2	372	No protein	Processed transcript	<u>.</u>		
Cab39l-203	ENSMUST00000224281.2	3546	No protein	Retained intron	5		

The strategy is based on the design of *Cab391-201* transcript, the transcription is shown below:



Genomic location distribution





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Protein domain

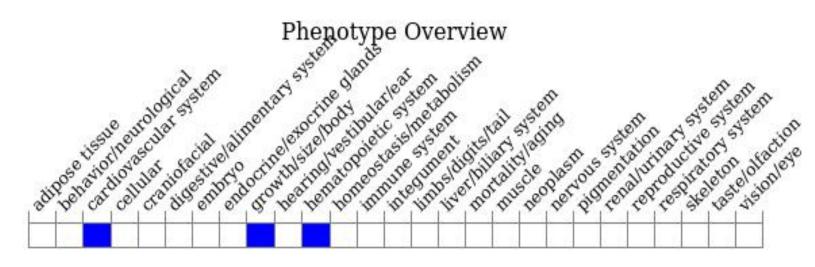


Superfamily Pfam	Arm Mo25-	adillo-type fold like							
PANTHER	Mo25-II	(e 182:SF9							
Gene3D	Arr	nadillo-like helic							
All sequence SNPs/i	Sequer	ice variants (dt	SNP and all o	ther sources)				í.	1 1
Variant Legend	sy	nonymous var	iant						
Scale bar	0	40	80	120	160	200	240	280	337

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Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).



If you have any questions, you are welcome to inquire. Tel: 400-9660890



